Genome-Wide Temporal Profiling of Transcriptome and Open Chromatin of Early Cardiomyocyte Differentiation Derived From hiPSCs and hESCs.

Public Summary:

Recent advances have improved our ability to generate cardiomyocytes from human induced pluripotent stem cells (hiPSCs) and human embryonic stem cells (hESCs). However, our understanding of the transcriptional regulatory networks underlying early stages (ie, from mesoderm to cardiac mesoderm) of cardiomyocyte differentiation remains limited. OBJECTIVE: To characterize transcriptome and chromatin accessibility during early cardiomyocyte differentiation from hiPSCs and hESCs. METHODS AND RESULTS: We profiled the temporal changes in transcriptome and chromatin accessibility at genome-wide levels during cardiomyocyte differentiation derived from 2 hiPSC lines and 2 hESC lines at 4 stages: pluripotent stem cells, mesoderm, cardiac mesoderm, and differentiated cardiomyocytes. Overall, RNA sequencing analysis revealed that transcriptomes during early cardiomyocyte differentiation were highly concordant between hiPSCs and hESCs, and clustering of 4 cell lines within each time point demonstrated that changes in genome-wide chromatin accessibility were similar across hiPSC and hESC cell lines. Weighted gene co-expression network analysis (WGCNA) identified several modules that were strongly correlated with different stages of cardiomyocyte differentiation. Several novel genes were identified with high weighted connectivity within modules and exhibited coexpression patterns with other genes, including noncoding RNA LINC01124 and uncharacterized RNA AK127400 in the module related to the mesoderm stage; E-box-binding homeobox 1 (ZEB1) in the module correlated with postcardiac mesoderm. We further demonstrated that ZEB1 is required for early cardiomyocyte differentiation. In addition, based on integrative analysis of both WGCNA and transcription factor motif enrichment analysis, we determined numerous transcription factors likely to play important roles at different stages during cardiomyocyte differentiation, such as T and eomesodermin (EOMES; mesoderm), lymphoid enhancer-binding factor 1 (LEF1) and mesoderm posterior BHLH transcription factor 1 (MESP1; from mesoderm to cardiac mesoderm), meis homeobox 1 (MEIS1) and GATA-binding protein 4 (GATA4) (postcardiac mesoderm), JUN and FOS families, and MEIS2 (cardiomyocyte). CONCLUSIONS: Both hiPSCs and hESCs share similar transcriptional regulatory mechanisms underlying early cardiac differentiation, and our results have revealed transcriptional regulatory networks and new factors (eg, ZEB1) controlling early stages of cardiomyocyte differentiation.

Scientific Abstract:

RATIONALE: Recent advances have improved our ability to generate cardiomyocytes from human induced pluripotent stem cells (hiPSCs) and human embryonic stem cells (hESCs). However, our understanding of the transcriptional regulatory networks underlying early stages (ie, from mesoderm to cardiac mesoderm) of cardiomyocyte differentiation remains limited. OBJECTIVE: To characterize transcriptome and chromatin accessibility during early cardiomyocyte differentiation from hiPSCs and hESCs. METHODS AND RESULTS: We profiled the temporal changes in transcriptome and chromatin accessibility at genome-wide levels during cardiomyocyte differentiation derived from 2 hiPSC lines and 2 hESC lines at 4 stages: pluripotent stem cells, mesoderm, cardiac mesoderm, and differentiated cardiomyocytes. Overall, RNA sequencing analysis revealed that transcriptomes during early cardiomyocyte differentiation were highly concordant between hiPSCs and hESCs, and clustering of 4 cell lines within each time point demonstrated that changes in genome-wide chromatin accessibility were similar across hiPSC and hESC cell lines. Weighted gene co-expression network analysis (WGCNA) identified several modules that were strongly correlated with different stages of cardiomyocyte differentiation. Several novel genes were identified with high weighted connectivity within modules and exhibited coexpression patterns with other genes, including noncoding RNA LINC01124 and uncharacterized RNA AK127400 in the module related to the mesoderm stage; E-box-binding homeobox 1 (ZEB1) in the module correlated with postcardiac mesoderm. We further demonstrated that ZEB1 is required for early cardiomyocyte differentiation. In addition, based on integrative analysis of both WGCNA and transcription factor motif enrichment analysis, we determined numerous transcription factors likely to play important roles at different stages during

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